

Continuous Fermentation and Stripping of Ethanol[†]

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Recycling the contents of a continuous fermentor through a stripping column is proposed as a means of reducing product inhibition and lowering the cost of fuel ethanol production. A 2-L fermentor and 10-cm packed column were continuously operated for 150 days without contamination. Some fouling of the packing with attached yeast cells was observed which partially blocked the column. Cell yield was lower than in a simple continuous fermentor. Complete conversion of 200 g/L glucose feed and 90% conversion of 600 g/L glucose feed were achieved. Data were analyzed by computerized process simulation. Cost analysis indicated that, with heat recovery to reduce heating and cooling costs, the continuous fermentor/stripper is possibly a lower-cost alternative to conventional fermentation and distillation.

Introduction

Production of ethanol by fermentation in simple batch or continuous fermentors is limited by the toxicity of the product. Because the fermenting microorganism, usually *Saccharomyces cerevisiae*, cannot tolerate more than about 10–12% by volume of ethanol, it is necessary to start with a relatively dilute glucose solution, usually not more than about 16% by weight, in order to achieve complete conversion in a reasonable time. The large amount of water carried through the process results in high costs for large process equipment for fermentation and subsequent purification of ethanol by distillation, which also requires the burning of a lot of fuel for steam.

It has been recognized for at least 20 years that there is the potential for considerable reduction in the cost of producing ethanol if ethanol separation is combined with fermentation. When ethanol is removed directly from the fermentor, or by recycling the contents of a continuous fermentor through a separation device which retains cell viability, it is possible to completely convert a much more concentrated glucose feed. With less water to carry through the process, and less to remove from the product, overall costs should be reduced.

Devices and methods for combined fermentation and ethanol separation have been recently reviewed (Park and Geng, 1992). These include fermentation under vacuum (Cysewski and Wilke, 1977), pervaporation (Müller and Pons, 1991; Shabtai et al., 1991), liquid-liquid extraction (Matsumura and Märkl, 1984; Kühn, 1980), perstraction (Christen et al., 1990), and solid adsorbents (Lencki et al., 1983). Biostil (Weatherly, Inc., Atlanta, GA) is a commercial process in which the contents of a continuous fermentor are recycled first through centrifuges to recover the yeast and then through a distillation column to remove ethanol. Bubbling of carbon dioxide gas through the fermentor to remove ethanol by stripping has been proposed (Walsh et al., 1983; Pham et al., 1989). It was calculated that the energy requirement to compress the gas 70 kPa for sparging into the fermentor was almost as much as the energy for distillation (Walsh et al., 1983). Stripping

from a packed column of immobilized cells was proposed (Dale et al., 1985). To overcome the high energy cost of compressing the stripping gas, while avoiding the need for immobilizing the yeast, it is now proposed to recycle the contents of a continuous fermentor through an ordinary packed column, where ethanol stripping with carbon dioxide could take place. Presented here are initial results from continuous operation of this system, along with computer simulation results and cost analysis.

Materials and Methods

Yeast. The yeast ATCC 4126, *Saccharomyces cerevisiae*, reported to have a high temperature tolerance and used in the vacuum fermentation work (Cysewski and Wilke, 1977), was obtained from the American Type Culture Collection, Rockville, MD. It was maintained on YM agar slants (YM agar, YM broth, and yeast extract were from Difco, Detroit, MI), sealed, and kept in the refrigerator for up to 1 year. The contents of one slant were resuspended in 100 mL of YM broth, grown in a 250-mL shake flask at 25–30 °C overnight and used to inoculate the fermentor.

Media. The fermentation medium for the initial experiments in the simple continuous fermentor consisted of 100 g/L glucose (dextrose, dry hydrate, 91.5% glucose, Corn Products Co., Inglewood Cliffs, NJ), 8.5 g/L yeast extract, 1.3 g/L NH_4Cl , 0.12 g/L $\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$, and 0.06 g/L CaCl_2 in tap water (Cysewski and Wilke, 1977). Glucose was sterilized separately. In later experiments in the simple continuous fermentor, 20 g/L corn steepwater (Grain Processing Corp., Muscatine, IA) was substituted for yeast extract and NH_4Cl was eliminated. Corn steepwater was sterilized, centrifuged to remove solids, and adjusted to pH 7.0 with NH_4OH before being added to sterile glucose and salts. For 200 g/L glucose media, all nutrient and salt concentrations were doubled. Due to a systematic error in media preparation, the 100 g/L glucose media were actually 94 g/L and the 200 g/L glucose media for the simple continuous fermentor were actually 194 g/L. For the continuous fermentor/stripper, the medium was the same except that the pH of corn steepwater was not adjusted and 0.067 g/L EDTA (ethylenediaminetetraacetic acid) was added to the 200 g/L glucose media to improve the clarity of the centrifuged corn steepwater media. EDTA was reported to improve the ethanol production in molasses media (Pandey and

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[†] Mention of brand or firm names does not constitute an endorsement by the U.S. Department of Agriculture over others of a similar nature.

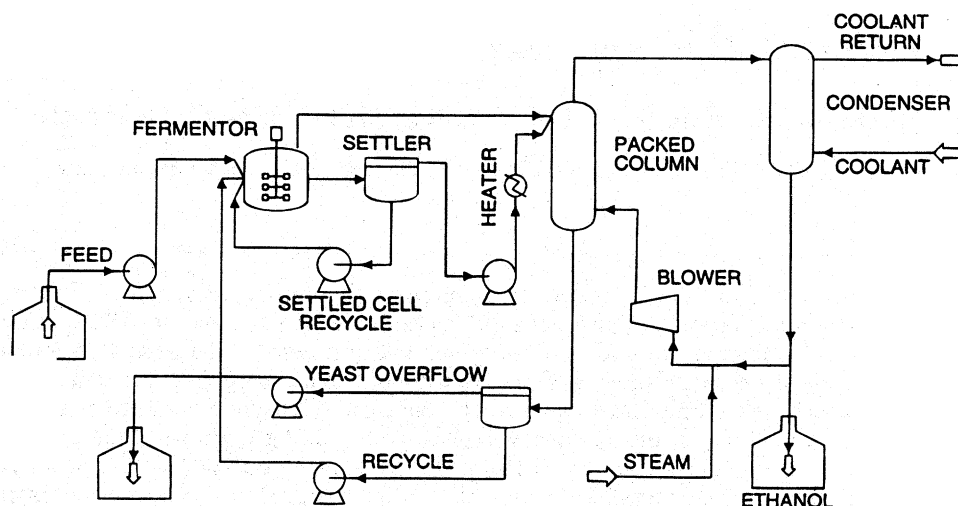


Figure 1.

Agarwal, 1993). For 400 or 600 g/L glucose media, all nutrient and salt concentrations were doubled or tripled.

Apparatus. The fermentor was a 2-L jar fermentor with gravity overflow to maintain the level at 1.2 L. In the simple continuous fermentor, the pH was continuously measured but not controlled. In the continuous fermentor/stripper, the pH was controlled at 3.75 by the automatic addition of 10% NH_4OH . Temperature and agitation were maintained at 35 °C and 300 RPM. The fermentor was continuously sparged with approximately 20 mL/min of air. Sterile feed solution was continuously pumped through an anti-grow-back tube and into the fermentor at various flow rates with a peristaltic pump. The actual flow rate was measured daily with the aid of a sterile buret on the feed line. In the simple continuous fermentor, the overflow drained by gravity into a sterile collection bottle. The drain line was equipped with an aseptic sampling line. In the continuous fermentor/stripper, the fermentor contents were recycled through a heater and into the top of a 10-cm-i.d. glass column packed to a height of 1.5 m with 4.4-cm plastic Tellerettes (Ceilcote Co., Berea, OH), as shown in Figure 1. The heater was a coil of 0.6-cm stainless tubing in a heating mantle controlled by a rheostat. The cell settler was a 250-mL separatory funnel, the cone-shaped section being 15 cm high and 7.5 cm wide at the top with the wall at a 15° angle from vertical. The cell settler and settled cell recycle pump shown in Figure 1 were not included in the initial setup but were added later. The small settler at the bottom of the column (25 mL) was intended mainly to ensure that the recycle pump pumped only liquid, while the yeast overflow pump pumped a mixture of gas and liquid, thus preventing any accumulation of liquid in the bottom of the column. Carbon dioxide was vented from the fermentor to the top of the packed column, accumulated in the gas recycle loop, and recycled by a blower through a glass-lined stainless steel jacketed condenser having inside diameter 7.6 cm and height 1.2 m. Carbon dioxide eventually escaped from the system around the blower drive shaft. The gauge pressure was less than 0.25 kPa throughout the gas loop. Coolant was supplied to the condenser jacket at -20 °C by two refrigerated baths. The condenser was packed with coiled, 1.0- × 20-cm strips of stainless steel to improve the mass and heat transfer. The cold gas leaving the condenser was reheated and humidified by direct steam injection. The temperatures of the gas entering the packed column and at various other key points around the system were measured and recorded hourly with thermistors and a data logger. The packed column,

condenser, and blower were connected by 10-cm glass and 7.6-cm stainless pipes, elbows, and tees. The condensate from the condenser drained by gravity through a U-tube trap to prevent gas entering or leaving and into a collection vessel. Steam condensate also drained through a U-tube trap. The packed column, condenser and coolant lines were insulated. The temperature in the packed column was manually controlled by adjustment of the steam injection needle valve and heater rheostat. Temperatures varied by several degrees, however, depending on room temperature and steam supply pressure. All components of the system that could not be sterilized in the autoclave were sterilized by flushing the system with nitrogen and recirculating 70% ethanol through the packed column with the blower on overnight. After the fermentor containing sterile medium was inoculated, the ethanol concentration was allowed to build up to about 40 g/L before the recycle pumps and refrigerated baths were turned on to prevent formation of ice in the condenser.

Analyses. Flow rates including the recycle pump flow rate, settled cell recycle pump flow rate (when present), yeast overflow rate, and condensate flow rate from the condenser were measured daily. The flow rate of steam condensate was measured weekly. Liquid samples were taken daily from the line between the fermentor and the packed column (after the settler when the settler was present) and from the bottom of the column (yeast overflow). Samples were analyzed for glucose, ethanol, and cell concentrations. A sample of the condensate from the condenser was also taken daily and analyzed for ethanol concentration. Cell concentration was measured by diluting the sample 1:20, measuring the optical absorbance at 600 nm, and comparing it to a standard curve of dry weights of yeast cells filtered on glass fiber filters and dried at 100 °C for 4 h. Glucose and ethanol were measured on the clear supernatant of the samples after centrifugation. Glucose was measured with an automatic glucose analyzer, Model 2000 (Yellow Springs Instruments, Yellow Springs, OH). Ethanol was measured with a gas chromatograph, Model 5890 (Hewlett Packard, Palo Alto, CA), with a flame ionization detector and SPB-1701 fused silica capillary column (Supelco, Bellefonte, PA). The oven program was 58 to 68 °C at 2 deg/min and 10 deg/min to 120 °C, which was held for 2 min. Ethanol was also measured on-line using a miniature gas chromatograph, Model M200 (Microsensor Technology, Fremont, CA) with OV-73 and OV-1701 columns and a thermal conductivity detector. The M200 was set up for continuous sampling with 4 h between

Table 1. Summary of Data from Simple Continuous Fermentor

(0)	(1) medium	(2) glucose (g/L)	(3) μ^a (h ⁻¹)	(4) cells (g/L)	(5) glucose (g/L)	(6) ethanol (g/L)	(7) pH	(8) yield (P/S)	(9) yield (X/S)	(10) conversion (g/h)	(11) conversion (%)
1	YE ^b	94	0.1010	7.0	0.55						
2	YE	94	0.1020	6.7	0.80	43.6	3.8	0.47	0.075	11.3	99.4
3	YE	94	0.1020	7.9	0.10	44.3	3.6	0.48	0.072	11.4	99.1
4	YE	94	0.1140	7.0	3.70	43.8	3.4	0.47	0.084	11.5	99.9
5	CS ^c	94	0.0760	5.2	0.44	42.9	3.9	0.48	0.078	12.4	96.1
6	CS	94	0.0900	4.9	3.20	45.5	3.4	0.49	0.056	8.5	99.5
7	CS	94	0.1010	4.8	9.26	46.6	3.4	0.51	0.054	9.8	96.6
8	CS	94	0.1020	6.4	0.60	41.3	3.4	0.49	0.057	10.3	90.1
9	CS	94	0.1020	5.1	3.76	42.7	3.8	0.46	0.069	11.4	99.4
10	CS	94	0.1020	4.7	10.60	44.7	3.4	0.50	0.057	11.0	96.0
11	CS	94	0.1020	6.6	0.80	39.8	3.3	0.48	0.056	10.2	88.7
12	CS	94	0.1050	5.2	3.00	44.5	3.6	0.48	0.071	11.4	99.1
13	CS	94	0.1050	5.4	2.80	46.8	3.5	0.51	0.057	11.5	96.8
14	CS	94	0.1050	4.5	7.60	48.0	3.5	0.53	0.059	11.5	97.0
15	CS	94	0.1180	5.2	7.90	41.5	3.4	0.48	0.052	10.9	91.9
16	CS	94	0.1320	5.0	11.20	44.7	3.5	0.52	0.060	12.2	91.6
17	CS	94	0.1320	4.3	13.80	41.9	3.4	0.51	0.060	13.1	88.1
18	CS	94	0.1470	4.2	21.00	38.7	3.3	0.48	0.054	12.7	85.3
19	CS	94	0.1600	3.9	25.00	35.4	3.4	0.49	0.058	12.9	77.7
20	CS	94	0.2160	3.4	39.40	34.4	3.4	0.50	0.057	13.2	73.4
21	CS	194	0.0242	2.6	39.30	25.1	3.6	0.46	0.062	14.2	58.1
22	CS	194	0.0375	3.1	47.00	78.3	4.3	0.51	0.017	4.5	79.7
23	CS	194	0.0503	4.1	56.20	70.4	4.3	0.48	0.021	6.6	75.8
24	CS	194	0.0640	3.5	67.70	65.4	4.2	0.47	0.030	8.3	71.0
25	CS	194	0.0760	4.2	72.00	63.3	4.0	0.50	0.028	9.7	65.1
26	CS	194	0.0890	4.0	70.80	65.4	4.1	0.54	0.034	11.1	62.9
27	CS	194	0.1150	4.3	87.70	62.5	4.0	0.51	0.033	13.2	63.5
						52.2	3.9	0.49	0.040	14.7	54.8

^a μ = specific growth rate = dilution rate. ^b Yeast extract. ^c Corn steepwater.

injections. The sample was a stream of nitrogen that was continuously equilibrated with the fermentor contents in a gas-liquid contactor. The gas-liquid contactor consisted of a length of silicone tubing inserted in a larger diameter polyurethane tube. The fermentor contents were continuously recycled through the silicone tube with a peristaltic pump while nitrogen was passed through the annular space between the silicone and polyurethane tubes. Equilibration occurred due to the permeability of silicone to ethanol vapor. The gas-liquid contactor was kept in a water bath at below room temperature to prevent condensation in the line to the M200. Results reported here include only those data obtained after sufficient time for the system to come to equilibrium. This was at least 1 day and usually several days after any change in operating variables for the simple continuous fermentor, and at least several days and usually 1 week after any change for the continuous fermentor/stripper. The ethanol data for the simple continuous fermentor reported here are averages of 4 or more data points from the on-line analysis. The ethanol data for the continuous fermentor/stripper reported here are from analysis of liquid samples, usually the average of two consecutive days' data.

Computer simulation and cost analysis were performed with the aid of Aspen Plus with Bioprocess Simulator (Aspen Technology, Cambridge, MA). The packed column was modeled as a single equilibrium stage with fermentation. Simple continuous fermentor data were fit to a kinetic model which included an empirical exponential dependence on ethanol concentration and the traditional Monod model for substrate dependence (Aiba et al., 1968). Process variables that were measured and input to the simulator included the glucose concentration and flow rate of the feed, the glucose and cell concentrations of the stream from the fermentor (or cell settler when present) to the packed column, the glucose and cell concentrations and flow rate of the yeast overflow, the ethanol concentration and flow rate of the condensate from the condenser, the recycle flow rate and settled cell

recycle flow rate (when present), the temperature of the gas entering the packed column and the flow rate of steam condensate. From these inputs, along with other constants supplied to the program such as the fermentor volume (1.2 L), ethanol yield (0.49), and ethanol inhibition constant (0.033 L/g) as determined from simple continuous fermentor data, the simulator calculated all other process variables including the gas flow rate, the temperatures of gas leaving the packed column and leaving the condenser, the effectiveness of the cell settler, the cell concentration and maximum specific growth rate in the fermentor, the glucose conversion rates in the fermentor and in the packed column, and the overall glucose conversion rate and percent conversion. The ethanol concentrations from the two liquid sample points containing cells (before and after the column) were measured but not used as simulation inputs. The agreement of these two measured values with the corresponding calculated values was an indication of the validity of the computer model.

Results and Discussion

Continuous Fermentor. Table 1 shows the results of experiments in the simple continuous fermentor. According to continuous fermentor theory, the dilution rate (feed flow rate divided by fermentor volume) is equal to the specific growth rate, μ , defined by

$$\frac{dX}{dt} = \mu X$$

where X is the cell mass. This assumes only that the flow rate leaving the fermentor is equal to the feed flow rate, a close approximation. As shown in Table 1, the specific growth rate varied from 0.0242 to 0.216 h⁻¹ in these experiments.

The specific growth rate is dependent on substrate (glucose) concentration, and agreement is generally found with the classical model.

$$\mu = \mu_{\max} \frac{S}{K + S}$$

where μ_{\max} is the maximum specific growth rate, S is the substrate concentration in the fermentor (equal to the substrate concentration in the effluent from a continuous fermentor), and μ_{\max} and K are kinetic parameters to be determined experimentally. In the case of ethanol fermentation, the specific growth rate also depends on the product (ethanol) concentration. An empirical exponential model has been found to agree well with data up to 60 g/L ethanol (Aiba et al., 1968). The complete model is thus

$$\mu = \mu_{\max} e^{-kp} \frac{S}{K + S}$$

where p is product concentration and k is a third kinetic parameter to be determined experimentally. Data for corn steepwater media containing 94 and 194 g/L glucose were fit to this model by nonlinear least squares. The results were $\mu_{\max} = 0.51 \text{ h}^{-1}$, $k = 0.033 \text{ L/g}$, and $K = 0.28 \text{ g/L}$. The model agreed well with the data up to about 60 g/L ethanol. At higher ethanol concentrations, the actual specific growth rate was less than predicted by the model, indicating that ethanol toxicity increases even more rapidly than the model predicts above 60 g/L.

In addition to the specific growth rate, two other parameters are needed to completely specify the fermentation kinetics. These are the cell and product yield factors, $Y_{X/S}$ and $Y_{P/S}$. These are defined as the grams of dry cells or ethanol produced per gram of glucose consumed or, in Table 1, column 8 = column 6/(column 2 - column 5) and column 9 = column 4/(column 2 - column 5). As shown in Table 1, the ethanol yield factor remained constant (within experimental error) at about 0.49 g of ethanol/g of glucose, while the cell yield factor varied from 0.017 to 0.084 g of cells/g of glucose. Cell yields were higher in yeast extract media than in corn steepwater media. Within the corn steepwater media data, the cell yield decreased with increasing ethanol concentration. Corn steepwater data were fit by linear regression and found to agree with

$$Y_{X/S} = 0.090 - 0.00095p$$

where p is ethanol concentration in grams per liter. In considering Table 1, the observation that is perhaps most significant for this work is that, while 94 g/L glucose feed could be completely converted, 194 g/L glucose feed could not be completely converted even at the lowest dilution rate.

Continuous Fermentor/Stripper. The analysis of the continuous fermentor/stripper data is not nearly as straightforward as for the simple continuous fermentor. While the overall glucose conversion rate and cell yield can still be derived from simple calculation on the yeast overflow data, most of the process variables are interrelated in a complex way and dependent on the highly nonideal vapor-liquid equilibrium for ethanol and water. The mass and energy balance equations for this system require trial-and-error solutions, and computerized process simulation is highly desirable if not essential. Table 2 shows data and simulation results. There were no data for the cell recycle flow (column 8) or the settler effectiveness (column 22) until after the continuous cell settler was added to the system. The only data shown in Table 2 that were not inputs to or results from the simulator are the ethanol data for the streams entering and leaving the packed column (columns 4 and 11). The agreement

of these data with the corresponding simulation results (columns 5 and 12) indicates that in most cases the internal consistency of the data and the validity of the computer model were quite good. Because the packed column was modeled as a single equilibrium stage, it can be further inferred from this agreement that the height of a theoretical mass transfer unit was approximately equal to the height of the column (1.5 m).

At the time of writing, the continuous fermentor/stripper had been operating continuously for 150 days without contamination. In contrast to the simple continuous fermentor, the continuous fermentor/stripper could easily convert 200 g/L glucose feed completely, and conversion of over 95% of 400 g/L glucose feed and over 90% of 600 g/L glucose feed were achieved. Approximately two-thirds of the overall glucose conversion took place in the packed column and one-third in the fermentor. The conversion in the fermentor was about 50% higher than in the simple continuous fermentor.

The gas flow rate dropped to about two-thirds of its initial value after the first few weeks of operation. This was due to accumulation of attached growth fouling the packing and partially blocking the column. Subsequent continuous operation did not result in further significant decrease of the gas flow rate, indicating that additional attachment or growth of attached cells was balanced by sloughing of attached growth from the packing. After several weeks of operation, clumps of aggregated yeast cells were observed which settled rapidly from the samples. Because the strain of yeast was nonflocculating, it was assumed that the clumps were derived from attached growth that was sloughed from the packing.

When the glucose in the feed was switched from 200 to 400 g/L, it was necessary to decrease the feed rate in order to maintain a high percent of conversion. This result did not agree with computer predictions. Also, the cell concentration in the fermentor was less than predicted. Cell yield, $Y_{X/S}$, was generally less than 0.02. The results from the simple continuous fermentor suggested that, at 50–60 g/L ethanol, the cell yield should have been 0.04–0.05. It thus seems likely that viable cells were being lost on passage through the column, possibly by settling out in stagnant spots in the column. It was decided to take advantage of the settling of yeast cells to increase the cell mass in the fermentor. When the glucose in the feed was increased to 600 g/L, the continuous settler was added between the fermentor and the column. The effectiveness of the settler, defined as the cell concentration in the concentrated stream divided by the cell concentration in the clarified stream, is shown in column 22 of Table 2. The cell concentration in the fermentor (now a simulation result) did increase to as much as 50 g/L, but the rate of glucose conversion in the fermentor remained about the same. This indicates that the settled cells being concentrated in the fermentor were not as active or were perhaps mostly dead. The low cell activity was also reflected in the maximum specific growth rate, μ_{\max} (column 24), calculated by the simulator, which decreased at the same time and almost in proportion to the increased cell concentration. Another indicator of cell death was to be found in the pH controller. Without the cell settler, base addition was needed to keep the pH at 3.75. With the cell settler, the pH drifted around 3.8–4.0 without any base addition. In batch culture, a rising pH usually accompanies cell death and autolysis, so it can be inferred that in this continuous system death and autolysis of settled cells were maintaining the pH.

During continuous operation with 600 g/L glucose feed there was some experimentation with varying the recycle

Table 2. Summary of Continuous Fermentor/Stripper Data and Simulation Results

(0)	feed		before column					(8) cell recycle (mL/min)	yeast overflow		
	(1) glucose (g/L)	(2) mL/min	(3) glucose (g/L)	(4) ethanol (g/L)	(5) ethanol ^a (g/L)	(6) cells (g/L)	(7) recycle (mL/min)		(9) mL/min	(10) glucose (g/L)	(11) ethanol (g/L)
1	200	2.43	26.8	26.4	36.0	4.7	4.2		2.75	0.4	15.6
2	400	1.92	51.8	51.7	55.2	8.3	9.6		2.01	16.7	47.7
3	400	1.94	79.9	52.1	53.1	6.2	7.1		2.17	30.1	52.8
4	400	2.22	63.3	46.2	50.7	6.5	9.7		2.32	14.6	46.9
5	400	2.28	60.9	45.9	52.7	7.2	9.9		2.20	13.9	42.9
6	600	1.42	133.0	51.6	49.5	4.9	16.8	11.4	1.56	108.0	50.6
7	600	1.46	77.8	51.0	50.7	8.9	33.1	11.6	1.22	62.1	48.6
8	600	1.47	72.0	57.7	58.9	7.7	24.8	11.2	1.59	55.2	55.4
9	600	1.50	82.5	58.6	60.1	8.7	33.4	11.7	1.47	68.9	56.6
10	600	1.51	95.9	47.3	51.1	9.0	36.9	34.3	1.07	82.1	46.9
11	600	1.53	111.5	52.3	50.1	5.9	25.1	10.9	1.68	90.4	52.8
12	600	1.61	115.1	51.6	56.0	8.6	23.0	22.9	1.38	93.8	48.0

(0)	yeast overflow		condenser condensate		steam condensate		gas				
	(12) ethanol ^a (g/L)	(13) cells (g/L)	(14) mL/min	(15) ethanol (g/L)	(16) mL/min	(17) ethanol ^a (g/L)	(18) kg/h ^a	(19) in temp (°C)	(20) out temp ^a (°C)	(21) cond ^a (°C)	(22) settler effect
1	21.7	3.1	0.777	186.0	10.6	3.1	2.22	39.2	29.3	-12.6	
2	50.7	7.8	0.595	337.0	11.3	4.9	1.52	37.7	26.9	-8.7	
3	51.0	4.9	0.550	324.0	11.2	5.2	1.74	37.8	24.1	-8.7	
4	49.6	6.3	0.727	318.0	11.2	6.2	1.77	38.7	28.5	-6.4	
5	50.8	6.7	0.747	320.0	10.6	7.9	1.61	37.3	30.5	-3.7	
6	47.7	4.0	0.593	330.6	18.0	3.4	1.51	39.2	27.1	-9.9	25.47
7	49.7	7.3	0.732	329.2	18.2	5.0	1.44	35.0	31.6	-5.3	12.80
8	56.2	5.6	0.652	341.1	11.7	6.2	1.73	38.2	26.6	-6.0	16.55
9	58.9	6.8	0.592	341.2	18.1	5.6	1.61	35.6	26.4	-4.6	13.20
10	50.0	8.2	0.752	337.9	17.6	5.2	1.27	30.6	33.7	-5.0	4.06
11	49.6	4.6	0.704	336.8	11.6	4.8	1.72	39.3	27.7	-9.0	22.00
12	54.3	7.3	0.684	327.4	17.5	6.4	1.50	34.1	30.3	-2.7	6.70

(0)	fermentor			column		overall		
	(23) cells (g/L)	(24) μ_{\max}^a (h ⁻¹)	(25) convn ^a (g/h)	(26) convn ^a (g/h)	(27) convn ^a (g/h)	(28) convn ^a (%)	(29) yield ^a (X/S)	
1	4.7	0.545	18.5	10.6	29.1	99.8	0.0175	
2	8.3	0.623	19.7	24.4	44.1	95.6	0.0213	
3	6.2	0.612	15.4	27.2	42.6	91.6	0.0149	
4	6.5	0.555	16.0	35.2	51.2	96.2	0.0171	
5	7.2	0.609	18.1	34.8	52.9	96.6	0.0167	
6	50.0 ^a	0.058	13.6	27.4	41.0	80.2	0.0091	
7	35.0 ^a	0.091	14.4	33.8	48.1	91.4	0.0110	
8	43.0 ^a	0.141	20.8	26.8	47.7	90.1	0.0112	
9	35.0 ^a	0.159	18.2	29.6	48.0	88.8	0.0124	
10	22.0 ^a	0.155	15.0	34.0	49.1	90.4	0.0107	
11	41.0 ^a	0.064	12.2	34.0	46.0	83.5	0.0100	
12	32.0 ^a	0.140	17.0	33.4	50.3	86.7	0.0120	

^a Simulation result.

and settled cell recycle flow rates. Although the data are not conclusive, it seems that, while the effectiveness of the cell settler and the cell concentration in the fermentor were reduced at higher recycle rates, the overall performance as judged by glucose conversion rate was improved. In particular, the glucose conversion in the column was increased at higher recycle rates. It is likely that there was better liquid distribution at higher recycle rates, allowing glucose to reach more of the attached cells, resulting in greater conversion.

Cost Analysis

For comparison purposes, it was first determined that, in most currently operating ethanol plants, the capital and operating costs attributable to fermentation and distillation amount to about \$0.05 per liter of anhydrous ethanol produced (Grethlein and Nelson, 1992). Then the cost was analyzed for the simplest continuous fermentor/strippler process including a continuous fermentor, packed

column, condenser, refrigeration unit, and distillation to anhydrous ethanol. It was found that the capital cost of just the continuous fermentor (\$270,000) was greatly reduced compared to the cost of batch fermentors (\$4,100,000). Distillation costs were also significantly reduced. The cost of the packed column, assuming performance was not affected by attached growth (\$910,000), was more than the fermentor, but still affordable. The biggest cost was for the condenser and refrigeration unit, which pushed the overall cost above that of current practice. However, it was found that, of the total heat removal duty on the condenser, one-half was for condensing the water vapor and one-sixth was for cooling the CO₂ gas. Only one-third was for condensing the ethanol vapor. The other two-thirds was fully recoverable. That is, heat exchangers could be employed to recover up to two-thirds of the heat removed from the warm, vapor-laden stream leaving the packed column and use it to reduce the steam requirement for reheating

and humidifying the cold dry gas leaving the condenser. After designing a network of heat exchangers to accomplish this, the overall cost including distillation to anhydrous ethanol was reduced to about the same as that of current practice, about \$0.05 per liter of product.

We believe that further improvements in the heat integration design are possible and that the continuous fermentor/stripper is possibly a lower-cost alternative to conventional fermentation and distillation. The major remaining uncertainty, and the one we are testing through experiments, initial results of which have been presented here, is the assumption that the performance of the column is not affected by attachment of yeast cells to the packing. On the basis of that assumption, the 10-cm column in our experimental setup should be able to strip about 50 L/day of ethanol. That would require a continuous fermentor of at least 50 L. So far, our experimental apparatus has produced no more than about 1 L/day of ethanol. However, we still have a very low pressure drop through the column. In the future, we will increase the fermentor size and blower speed until we reach the flooding point of the column. From the observed effects of attached growth reported here, it seems likely that flooding will occur before the 50 L/day theoretical performance of clean packing is reached. However, the packed column is not the major cost in the process, and if it must be several times larger than assumed, corresponding to perhaps 10 or 20 L/day of ethanol stripped in the 10-cm column, then the overall cost could still very possibly be less than that of current practice.

Conclusion

Recycling the contents of a continuous fermentor through a stripping column reduces product inhibition and permits high conversion of a concentrated glucose feed, which is not possible in a simple continuous or batch fermentor. Continuous operation of a 2-L fermentor and 10-cm packed column for 150 days without contamination and 90% conversion of 600 g/L glucose were achieved. Some fouling and blockage of the packed column with attached growth of yeast cells was observed, but the column exhibited stable performance at a stripping rate of about 1 L/day of ethanol. If, in future experiments with a larger fermentor, 10 L/day of ethanol or more can be stripped in the 10-cm column, then a process based on continuous fermentation and stripping might be a cost-effective replacement for conventional batch fermentation and distillation. Such a development could provide significant benefits for corn growers, fuel ethanol producers, and vehicle operators.

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